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BRIEFER ARTICLES.

OBSERVATIONS UPON THE FEEDING PLASMODIA OF *FULIGO SEPTICA*.

(WITH ONE FIGURE)

ALTHOUGH considerable attention has been paid to the plasmodia of the Mycetozoa, especially by the German botanists (De Bary, Zopf, Sachs), little work has been done upon the feeding habits of these interesting protoplasmic masses. In a valuable contribution to the life history of these organisms, Lister¹ sets forth in a painstaking way the manner in which the plasmodium of *Badhamia utricularis* behaves when actively feeding. Various substances were tried by way of experiment. Pieces of *Agaricus campestris*, *A. melleus*, *A. rubescens*, *A. fascicularis*, *Boletus flavus*, and *Corticium puteanum* were used, but none of these fungi seemed so desirable a food as *Stereum hirsutum*, which was devoured without leaving any residue. *Agaricus fascicularis* was found in these experiments to be a particularly unwholesome morsel. The digestion by the active plasmodium of the fungi above mentioned presupposes the presence of a nitrogenous ferment, namely a proteo-hydrolytic one. As far as Lister's observations show, starch seems to be refused by the moving plasmodium, contradicting the idea of the presence of a diastatic ferment. The following observations upon the plasmodium of *Fuligo septica* is in part a contribution to the life history of plasmodia in general.

While searching for Mycetozoa in the wooded valley incorporated as part of Woodlands cemetery, West Philadelphia, a luxuriant growth of *Pleurotus sapidus* was found upon some partially decayed logs, which had been piled up in a loose manner preparatory to burning. In removing several large pieces of this fungus, small patches of yellow plasmodium were found upon the lamellar surface of the fully expanded pilei. These protoplasmic masses had moved out from the rotten log where they were seen in the crevices, and had invaded the gill surface

¹ Notes on the plasmodium of *Badhamia utricularis* and *Brefeldia maxima*. Ann. Bot. 2 : 1-23. 1888.

of *Pleurotus sapidus*. The appearance of the larger plasmodium at this time may be described as follows: The gills which were still rigid and in natural position were connected in the invaded portions of the lamellar surface by bridges of slimy yellow protoplasm. The basidial layers were covered by the more delicate portions of the plasmodial reticulum. The larger, more cord-like streams of protoplasm stretched from gill to gill, connecting as main cables the outlying pseudopodial fingers of protoplasm. The plasmodia growing upon several separate pieces of fungus were removed at 2 P. M. Friday, November 2, carried to the botanical laboratory of the University of Pennsylvania, and covered by two bell jars provided with dampened filter paper. By 6 P. M. of the same day the larger plasmodium had increased to such an extent as to cover completely the fungal pieces under one of the bell jars, and the gills showed signs of collapse. At 9 A. M., Saturday, November 3, the gills were found to be in a total state of collapse, Fuligo by this time having taken complete possession. Under the other bell jar the plasmodium, which was originally about the size of a silver dollar in superficial extent, had increased until it had spread to the outer circumferential margin of the lamellar area. In their attack upon the edible portions of *Pleurotus*, masses of protoplasm heaped themselves up into rounded knobs, or protuberances formed by condensations of the myxomycete substance. These would disappear, to be finally replaced by others of similar size and form. These observations were made on Saturday morning. The invasion and destruction of the gill surface was complete by Monday.

An examination of the growth under the bell jars showed a most remarkable development of the larger plasmodium. It not only covered the fungus, but also the inner sides of the bell jar in the form of a beautiful yellow reticulum. The wet filter paper plastered upon the top of the bell jar was completely covered by a dense mass of anastomosing protoplasm. Upon the main currents of plasmodial movement were beads of protoplasm of larger and smaller size. Where these hung, as pendent drops on the moist filter paper, they had grown until the protoplasm hung, as yellow stalactites, dangling from the dome-like roof of the inner side of the bell jar. The dome of the bell jar on Monday was almost entirely covered by the yellow plasmodium.

A strip of filter paper with the actively streaming plasmodium of Fuligo was removed from the moist chamber and placed in a dry situation in the bright sunlight. As the filter paper dried, the protoplasm

rapidly streamed to the wettest portion, and then began to aggregate into an extended aethalium. The drying, however, took place so rapidly that the entire plasmodium had not time to withdraw itself from the filter paper, and therefore it dried *in situ*, leaving a characteristic network of dry anastomosing threads. The reproduced photograph was taken by Mr. W. H. Walmsley just before the moving plasmodium was placed in the sunlight.

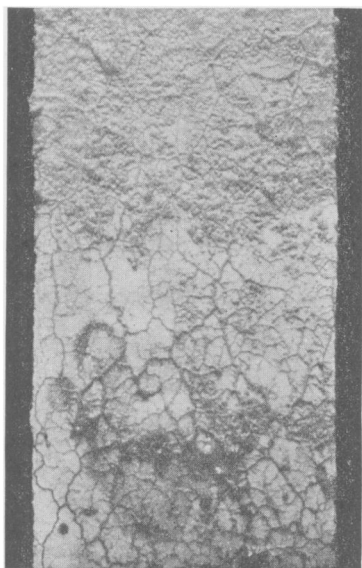


FIG. 1.—Moving plasmodium of *Fuligo septica* on moist filter paper, showing the varicose condition of the reticulum while actively streaming.

Beneath the bell jars, as the disintegration of the fungi proceeded with the production of a watery fluid and a gelatinous substance of a ropy consistency, the more active protoplasmic masses confined themselves to the tougher portions (the stipe and main substance of the pileus), heaping up on these portions in the concentrated effort to digest them. With the drying of the interior of the bell jars, condensation of the reticulum took place, so that the meshes became smaller and the anastomosing streams more closely aggregated. At this stage of growth experiments were begun by feeding *Fuligo* various substances.

At 1 : 30 P.M. Tuesday, November 6, pieces of fresh young *Pleurotus* were placed upon the living plasmodium. At 2 : 15 P.M. the small pieces used were invaded and the plasmodium had spread over about half of the superficial surface of the fungal food. Pieces of young pilei of *Coprinus comatus* were placed on different spots of the same plasmodium, and by 2 : 15 P.M. invasion had well advanced, but the rapidity of forward movement was much less when *Pleurotus* was used as a food. Two hours after the two food substances had been placed within reach of the streaming protoplasm, complete covering of the specimens had occurred.

Pieces of a partially dried toadstool, *Hypholoma perplexum*, were also

placed within contact of another plasmodium of *Fuligo* at 1:30 P.M., November 6, but at five o'clock not a single pseudopodial branch of the plasmodium had moved up upon them. On the contrary, when at four o'clock several pieces of the pileus and stem of *Coprinus atramentarius* were laid upon the yellow protoplasmic mass, inside of three quarters of an hour the trophotropic action of the food substance began to manifest itself by the plasmodial invasion of the newly provided nitrogenous food substance. The incorporation of the nutritive matter had well proceeded up over the edges of the young pilei of *Coprinus atramentarius* by five o'clock, November 6. An examination on the morning of November 7 showed the inky *Coprinus* almost entirely digested, and a black mass of spores in a gelatinous matrix indicated a total collapse of the fungus. *Hypholoma perplexum* was also covered by a network of the mycelium, which had spread not only over the stipe, but also over the gills and upper surface of the pileus. By evening, this agaric had collapsed, and by the next morning, November 8, nothing remained but a soft gelatinous mass of substance.

Raw beefsteak was applied to the surface of the plasmodium at 11:30 A.M., November 7, and by noon a single strand of protoplasm had advanced upon the meat. At 1:30 P.M. one third of the surface of the meat, and by 5:00 P.M. the entire surface, was covered. Digestion must have been rapid during the night, because upon returning to the laboratory in the morning of November 8 not a trace of the beefsteak was to be found.

Pieces of the gleba and stipe of *Phallus impudicus* were also applied at the same time. During the afternoon of Wednesday, November 7, the pieces of gleba were well covered by the moving plasmodium, the stipe portions being left untouched. By the next morning the glebal pieces had almost entirely disappeared, and cuts from the stipe still remained untouched.

Beefsteak was again supplied to the plasmodium at 10:00 A.M., Thursday, November 8, and by 10:45 A.M. a few arms of the plasmodium had extended themselves over the free edges of the meat.

The purpose was next to extend the series of observations by feeding to the active plasmodium a variety of nitrogenous and fatty materials. Cheese, boiled white of egg, boiled yolk of egg, and butter were chosen. Pieces of these substances were applied to the surface of the reticulum on Thursday morning, November 8. The plasmodium seemed at first to refuse them, but by Friday morning the fragments

of boiled white of egg were found to be partially covered by the creeping Fuligo. On Saturday morning, November 10, the hardened, coagulated egg albumen was completely covered and well-nigh digested. The yolk was but slightly affected by the plasmodium, even after exposure to the digestive action for two whole days. The butter was left untouched.

The presence of several ferments is naturally inferred from the digestive action accomplished by the plasmodium. According to De Bary, diastase can be extracted from the plasmodium of *Æthaliu* (*Fuligo*).²

In his book on ferments Green³ states: "One of the earliest known of these is the ferment which Krukenberg found to be procurable from the plasmodium of *Æthaliu*, one of the *Myxomycetes*. A glycerine extract of the plasmodium was found to have very marked proteolytic powers in the presence of lactic or hydrochloric acid. Krukenberg's statement has been confirmed by Miss Greenwood, who has stated that the plasmodium of another member of the same group yielded to 0.4 per cent. hydrochloric acid an extract which showed marked solvent action on fibrin." Negative results were obtained when I removed some of the partially digested fungus with plasmodium upon it, and treated the mass with glycerine, according to the directions given above. To the glycerine extract, which had a slightly yellowish color, a few drops of 35 per cent. hydrochloric acid was added, and a small frayed piece of raw beefsteak. After two days of trial the beefsteak was found unchanged, although left in the glycerine extract for that length of time.

The plasmodium brought into the laboratory on Friday, November 2, was still in a streaming condition on Saturday, November 10, when observation upon it ceased. The original fungus, with the exception of the more fibrous stipe, had in this time been reduced to a fibrous gelatinous mass, upon which the plasmodium still streamed, finding apparently enough remaining food to feed upon, although by this time the common mold had invaded it. This mold appeared for the first time on Wednesday, November 7, but was then brushed off to prevent fruiting. The plasmodium, while actively streaming and feeding, kept the substratum remarkably sweet and clean, and it was not until the original food substance had been destroyed as food that foreign organisms, such as the mold, had any chance for development. This

² This statement is somewhat at variance with the observations of Lister, *loc. cit.*

³ The soluble ferments and fermentation 215. 1899.

was certainly one of the most instructive facts brought out during the course of my observations.⁴—JOHN W. HARSHBERGER, *University of Pennsylvania*.

SWARM SPORE FORMATION IN HYDRODICTYON UTRICULATUM ROTH.⁵

1. The methods of fixing were by means of Merkel's fluid and a mixture of iridium chlorid and acetic acid according to one of the following formulæ:

(1) Eisen.	Iridium chlorid (0.5 per cent. aqueous solution)	-	-	-	-	-	-	100 ^{cc}
	Glacial acetic acid	-	-	-	-	-	-	1 ^{cc}
(2) Iridium chlorid	(1 per cent. aqueous solution)	-	-	-	-	-	-	100 ^{cc}
	Glacial acetic acid	-	-	-	-	-	-	3 ^{cc}

The best results were obtained with the stronger iridium chlorid mixture.

2. There is no differentiated chromatophore in the cell. The pyrenoids and nuclei are scattered irregularly throughout the cytoplasm and the chlorophyll is contained in the whole cytoplasmic body. The nuclei in both the resting and dividing stages show the structure typical of higher plants and are not to be taken as types of primitive nuclei.

3. Cleavage takes place by means of surface constriction of the plasma membrane on the outside and the vacuolar membrane on the inside of the protoplasmic layer. The process is a progressive one, the cleavage furrows cutting out first large irregular multinucleated masses of protoplasm, which are in turn divided into smaller masses, until each contains a single nucleus, the entire protoplast thus being divided into spores. The swarm spores are uninucleated biciliated cells. At the base of the pair of cilia there is a clearly defined basal body.

A detailed description of the processes outlined above will be published soon in a more complete form.—H. G. TIMBERLAKE, *University of Wisconsin*.

⁴ One of the best methods of procuring material for microscopic study is to remove the protoplasm by scraping, and then to place portions of it on slides fitted into the bottom of Petri dishes provided with moist filter paper. In an hour or two these mounds of protoplasm will have spread out over the slides sufficiently to permit of their examination. At my suggestion, Dr. Mazjick Ravenel, bacteriologist of the Pennsylvania Live Stock Sanitary Board, tried to grow the plasmodium upon ordinary agar, and upon filter paper saturated with bouillon, but failure resulted in both cases.

⁵ Résumé of results presented at the meeting of the Western Naturalists at the Hull Botanical Laboratory, December 27, 1900.